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TITLE

COLD-HARDINESS OF OVERWINTERING  
MOUNTAIN PINE BEETLE LARVAE

by  
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Coeur d'Alene, Idaho  
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October 2, 1941

To: Dr. F. C. Craighead, in Charge, Forest Insect  
Investigations

From: James C. Evenden, Box 630, Coeur d'Alene, Idaho

Subject: Report "Cold-Hardiness of Overwintering Mountain  
Pine Beetle Larvae"

I am enclosing copy of a report entitled "Cold-Hardiness of Overwintering Mountain Pine Beetle Larvae", which covers the tests conducted at this laboratory during the winters of 1939-40 and 1940-41.

You will note that we have changed our technique, and that instead of using exposed larvae in petri dishes we expose infested log sections, which permits us to obtain an accurate figure as to the mortality associated with each exposure.

Although it would seem that these data rather definitely establish the relationship between cold-hardiness and winter temperatures, I am planning to conduct a few additional tests during the coming season for the purpose of obtaining a check upon last winter's results.

From these data it is believed that by studying weather records during the fall of the year one might well hazard a prediction as to the cold-hardiness that will follow.

Your comments and suggestions concerning this report will be appreciated.

Copies are being sent to the Berkeley, Portland, and New Haven Laboratories.

James C. Evenden

Enclosure

**COLD-HARDINESS OF OVERWINTERING  
MOUNTAIN PINE BEETLE LARVAE**

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# COLD-HARDINESS OF OVERWINTERING MOUNTAIN PINE BEETLE LARVAE

by  
James C. Evenden  
Senior Entomologist

## INTRODUCTION

Studies conducted at the Coeur d'Alene Laboratory during the past five years have clarified the relationship which exists between winter temperatures and unusual mortalities of mountain pine beetle broods overwintering in white pine. Although no positive conclusions can be drawn as to the degree of mortality which will occur under specific temperatures, these studies have established those conditions from which beneficial mortalities can be expected.

Although the accuracy of the conclusions drawn from this accumulation of data are not questioned, it is realized that objections can be offered to the technique of removing larvae from infested logs for subsequent exposure to desired temperature in specially prepared petri dishes. These objections would rest in the unnatural condition of exposure and the physical disturbance which, regardless of all precautions, must have occurred. Perhaps the greatest objection would be the difficulty of determining actual brood mortalities, as larvae recorded as recovering from exposures could not be reared to maturity.



To meet this objection, experiments were planned to remove all doubt from the accuracy of the data, as well as to check previously drawn conclusions. Although unusual weather conditions influenced the results of these experiments as far as their complete objective was concerned, the results of the past two seasons' work are presented as an additional contribution toward a better understanding of low temperatures and bark beetle mortality.

#### EXPERIMENTS OF THE 1939-1940 SEASON

To measure under as near normal conditions as possible the cold-hardiness of overwintering mountain pine beetle broods at different periods in the fall and winter, the following plan was adopted:

(1) Logs 36 inches in length and varying from 13 to 16 inches in diameter were cut from infested trees in October 1939 and stored in an open shed under what was assumed to be normal forest conditions.

(2) At approximately two-week intervals during the winter one of these logs was exposed to a temperature which was reduced from a point comparable to air conditions of that day, to a subcortical temperature of  $-30^{\circ}$  F.

(3) Prior to exposure a 6-inch block was cut from each end of the 36-inch log, leaving a 24-inch section for exposure within the low-temperature cabinet. No attempt was made to protect the exposed log ends, as previous experiments had shown that effects of such an exposure could only be traced for an inch or two. As the examination

of treated logs was confined to the center 12 inches, a 6-inch protection strip was left on each end.

(4) An examination of the 6-inch blocks provided a figure as to the potential brood within the treated section.

(5) Exposed logs were stored in an open insectary, and examined after insect activity had started in the spring. Untreated logs were stored with them as a check upon these results.

Based upon previous data it was expected that in the fall a complete mortality would follow an exposure to this temperature, but during November considerable survival would occur, and by December sufficient cold-hardiness would prevail to prevent all mortality. It was also expected that with the return of activity temperatures in February or March, larval resistance would be broken and complete mortality would again be associated with this exposure. This procedure was employed to show the normal development of larval resistance to what has been assumed to be the extreme subcortical temperatures of the region, and to check actual mortality as indicated by previous experiments.

Although this plan seemed to satisfy all requirements of our problem as indicated by previous experiments, the rather unprecedented mild winter was reflected in a low degree of larval resistance. At no time during the winter was sufficient cold-hardiness developed to withstand the planned exposure to  $-30^{\circ}$  F., although this condition had prevailed <sup>during</sup> three previous seasons. As a result of this unforeseen condition the desired results were not obtained; however, the data show interesting facts and are presented in the following table:

Table 1 - Brood mortality in white pine logs exposed to a lowering cabinet temperature which produced a subcortical temperature of  $-30^{\circ}$  F.

Test	Date	Exposure	Living brood per sq. ft. Estimated brood per sq. ft. of bark surface at time of exposure	of bark surface at time of examination May 1940	Mortal- ity
1	10/1/39	38 hours From $+60$ to $-30.1^{\circ}$ F.	Larvae 63 Pupae 13 N. A. 28	104 0	100%
2	10/17/39	52 hours From $+50$ to $-30.6^{\circ}$ F.	Larvae 48 Pupae 5 N. A. 31	84 0	100%
3	10/31/39	50 hours From $+45$ to $-30.6^{\circ}$ F.	Larvae 31 Pupae 4 N.A. 24	59 0	100%
4	11/14/39	50 hours From $+45$ to $-30.1^{\circ}$ F.	Larvae 28 Pupae 2 N. A. 16	46 0	100%
5	12/3/39	61 hours From $+45$ to $-30.7^{\circ}$ F.	Larvae 51 Pupae 1 N. A. 19	Larvae .41 Pupae .13 71	99.4%
6	1/2/40	43 hours From $+40$ to $-30.3^{\circ}$ F.	Larvae 95 Pupae 0 N. A. 1	Larvae .28 Pupae .70 96	99.0%
7	2/5/40	40 hours From $+50$ to $-30^{\circ}$ F.	Larvae 52 Pupae 0 N. A. 0	Larvae .57 Pupae .71 52	97.6%

The data in table 1 show a much lower degree of cold-hardiness than existed during the three previous seasons. As a slight brood survival occurred in tests 5, 6, and 7, it can be assumed from a knowledge of resistance trends that from December to February a cold-hardiness prevailed which would have successfully withstood an exposure of approximately  $-20^{\circ}$  F. In connection with the February experiments (test No. 7) data were obtained from an exposure of naked larvae in petri dishes which indicated a low degree of cold-hardiness for that period. This information showed that a continuation of the planned experiments would be of little value, so the plan was discontinued. Additional exposures to higher temperatures were then made to obtain some measurement of the resistance which did exist. These data (table 2) show interesting relationships between different exposures and mortality.



Table 2 - Brood mortality in white pine logs exposed to a lowering temperature to produce subcortical temperature ranging from 0 to -25° F.

Test:	Date	Exposure	Estimated brood per sq. ft. of bark surface at time of exposure	Living brood per sq. ft. of bark surface at time of examination	Mortal- ity
				May 1940	
8	2/20/40	45 hours From +40 to -20.3° F.	Larvae 189 Pupae 0 N. A. 0	Larvae 39 Pupae 2 41	76.3%
9	3/4/40	35 hours From +30 to -25.5° F.	Larvae 94 Pupae 24 N. A. 0	Larvae 1.6 Pupae .58 2.18	98.2
10	3/6/40	47 hours From +40 to -21.3° F.	Larvae 137 Pupae 0 N. A. 0	Larvae 3.8 Pupae .25 4.05	97.1
11	3/12/40	26 hours From +31 to -14.6° F.	Larvae 244 Pupae 0 N. A. 0	Larvae 115 Pupae .51 115.51	52.7
12 A	4/16/40	21 hours From +35 to 0° F.	Larvae 197 Pupae 0 N. A. 0	Larvae 4.84 Pupae 0 N. A. 0	97.6
12 B	4/16/40	27 hours From +35 to -7.2° F.	Larvae 197 Pupae 0 N. A. 0	Larvae 0 Pupae 0 0	100
12 C	4/16/40	31 hours From +35 to -16.8° F.	Larvae 197 Pupae 0 N. A. 0	Larvae 0 Pupae 0 0	100

Activity temperatures have been shown\* to be lower for pupae than for larvae, so that when larval activity is checked by a drop in temperature, pupae are capable of completing their development to new adults, providing there are no abnormal weather conditions. As pupae were found in check logs as late as December 3, it is apparent that activity temperatures prevailed until that date. The exposure of infested logs under such conditions could be considered as an occurrence of an unseasonal temperature at a time when the larvae had not been subjected to temperatures sufficiently severe to develop adequate cold-hardiness. It is still further apparent that activity temperatures existed during the latter part of February and early March, as 20% of the brood had pupated on March 4. Activity temperatures are also evidenced by the lowered resistance shown by brood mortalities associated with tests 8 and 9.

The fact that the logs used in connection with tests 10, 11, and 12 all contained small larvae resulting from late fall attacks explains the absence of pupae. The same mortality is shown for tests 9 and 10, although there was a difference of 5° in the exposures. Test 9 was directed against broods of mature larvae, and test 10 against broods of small larvae, which are less resistant. Tests 11 and 12 show the rapidity with which larval resistance drops with the advent of warm weather, as severe mortality followed exposures to high temperatures.

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\* Temperature Requirements for Pupation of the Western Pine and Mountain Pine Beetles. J. E. Whiteside, May 20, 1935. Unpublished report.

At the time the treated logs were examined in May, a number of Coeloides dendroctini Cushman cocoons (parasites of the mountain pine beetle) were taken from logs where the beetle brood had been destroyed through an exposure to  $-30^{\circ}$  F., and placed in rearing jars. An emergence of 49 adults was obtained from 54 cocoons, with the time of emergence being comparable to that from untreated material. This fact offers a possibility that these parasites have a higher degree of low-temperature resistance than their host.

#### EXPERIMENTS FOR THE 1940-1941 SEASON

Experiments for the 1940-1941 season were planned to provide reasonable protection against such unusual winter conditions as previously experienced. Infested logs were used again with the same plan of controls, but instead of exposing one log to a definite temperature for each period, from four to six logs were used for each test, and exposed to a range of temperatures which were expected to measure maximum resistance for each period. The plan of storing exposed logs to outside temperatures until spring activity before examination was discontinued to avoid the possibility of results being influenced by other factors. The plan followed was to hold both the exposed logs and their checks at room temperatures for 10 to 14 days. An examination at that time showed without question the mortality associated with the treatment.

The general plan of experiments worked quite satisfactorily, but again the writer proved to be a rather poor prophet concerning the character of the winter and the degree of cold-hardiness developed by the larvae. In three instances the range of temperatures selected for the experiment were not adjusted so as to include the maximum resistance. The data obtained from these tests are shown in table 3:



Table 3 - Brood mortality in white pine logs exposed to lowering temperatures to produce different subcortical temperatures

Date		Exposure	Living of check log	Dead	Brood per sq. ft. of bark surface at time of examination	Living Dead	Mortality
Dec. 2-6 1940	I A	8 hours From +45 to +4.20° F.	Living 10.7 Dead 0		Living 13.1 Dead 0		0
	B	9 hours From +45 to +0.35° F.	Living 17.5 Dead 0		Living 15.6 Dead 0		0
	C	10 hours From +45 to -3.75° F.	Living 17.6 Dead 0		Living 19.9 Dead 0		0
	D	14 hours From +42 to -9.75° F.	Living 8.1 Dead 0		Living 15.0 Dead 0		0
	E	28 hours From +48 to -14.5° F.	Living 8.0 Dead 0		Living 4.0 Dead 5.3		57½
Jan. 6-12 1941	II A	22 hours From +32 to -49° F.	Data not recorded for A & B, as there was no mortality for II C				0
	B	24 hours From +32 to -10.2° F.					0
	C	23 hours From +32 to -16.3° F.	Living 39.4 Dead 0		Living 35.6 Dead 0		0
	D	26 hours From +32 to -20.8° F.	Living 38.1 Dead 0		Living 33.8 Dead 0		0
	E	26 hours From +32 to -25.0° F.	Living 42.7 Dead 0		Living 28.7 Dead 9.3		24.5
	F	32 hours From +32 to -30.1° F.	Living 46.7 Dead 0		Living 28.0 Dead 12.7		31.2
Feb. 3-7 1941	III A	27 hours From +32 to -19.8° F.	Living 31 Dead 0		Living 3.9 Dead 11.7		75
	B	29 hours From +32 to -25.0° F.	Living 31 Dead 0		Living 1.6 Dead 12.4		88
	C	34 hours From +32 to -29.1° F.	Living 9.4 Dead 0		Living .5 Dead 10		95
	D	49 hours From +32 to -34.6° F.	Living 9.4 Dead 0		Living .5 Dead 8.9		94
Feb. 13-19 1941	IV A	25 hours From +41 to -20.3° F.	Living 32 Dead 0		Living 47.2 Dead 12.2		21
	B	31 hours From +41 to -25.5° F.	Living 32 Dead 0		Living 16.7 Dead 25.9		61
	C	37 hours From +40 to -29.6° F.	Living 31.6 Dead 0		Living 0 Dead 38.8		100
	D	50 hours From +40 to -34.1° F.	Living 31.6 Dead 0		Living 0 Dead 40.0		100



Table 3 continued

:	:	:	:Brood per sq. ft.:			
:	:	:	:Brood per sq. ft.:of bark surface :			
:	:	:	:of bark surface		:at time of	:Mortal-
:	:	Exposure	:of check log		:examination	: ity
March	V A	27 hours	Living	35.2	Living	.58
17-20		From 46° to -10.2° F.	Dead	0	Dead	26.40 98%
1941	B	29 hours	Living	35.2	Living	0
		From 46° to -15.3° F.	Dead	0	Dead	35.2 100
	C	35 hours	Living	33.9	Living	0
		From 50° to -19.8° F.	Dead	0	Dead	26.1 100
	D	38 hours	Living	33.9	Living	0
		From 50° to -28.0° F.	Dead	0	Dead	28.4 100

The preceding data (table 3) show that during the winter of 1940-41 overwintering mountain pine beetle larvae developed sufficient cold-hardiness to withstand, in January, an exposure of -30° F. (subcortical) with but a 31% mortality. However, this resistance was subsequently broken, for a similar exposure in early February resulted in a 95% mortality, and in March an exposure to -15° F. resulted in 100% mortality. The breaking of this resistance in February was undoubtedly due to warm temperatures (chart IV) occurring at the time the tests were conducted.

As a check upon the technique being employed, exposed larvae in petri dishes as used in previous experiments were exposed in connection with the February test. These check tests consisted of two exposures: one of 8 hours from -25.5° F. to -29.1° F.; and one for five hours from -34.1° F. to 34.6° F. Four days after exposure the first test showed a brood mortality of 92.5%. Although in the second test the larvae were exposed to a lower temperature, the mortality was only 36.7%. A comparison of these figures with the results obtained from the exposed log would indicate a slightly higher degree of resistance for the petri dish larvae. However, it is quite difficult to determine the actual mortality associated with

petri dish exposures. Although larvae that are considered as having survived the severe exposures are active, and in many instances feed upon fresh phloem which is provided, we have been unable to rear them to maturity. Although these two techniques may offer a satisfactory measurement of cold-hardiness, the difficulty of determining actual mortality makes the use of infested logs a preferred procedure.

Although the winter of 1940-41 was generally considered as being abnormally mild, the high degree of resistance attained by the larvae during January 1941 was not at variance with the weather condition which occurred during the previous fall. A comparison of charts II and IV will show that the fall temperatures of 1940 were even more severe than those of 1938, which were considered as being responsible for the development of a high degree of cold-hardiness. As it is apparent that the character and severity of fall temperatures determine the degree of cold-hardiness developed, the reason for the larval resistance shown during January is quite obvious.

The exposure of infested logs with subcortical temperatures recorded by thermocouples, with an examination of the logs from 10 to 14 days later, provides a technique of operations as free from criticism as any procedure could be. In such an examination of exposed logs there are no border lines between larval death and recovery. Mortality is represented by dead or badly discolored larvae, the death of which can be assured. The resistant brood were active feeding larvae, with many pupae and new adults having developed since exposure. In no instance was there any question between recovery and death, nor could the cause of death be questioned.

SEASONAL RESISTANCE OF MOUNTAIN PINE BEETLE LARVAE  
WINTER 1938-1939

The results of experiments conducted during the winter of 1938-1939 are shown in chart I. These data were presented in a previous report\*, but are used again to illustrate the high degree of cold-hardiness developed by the mountain pine beetle during that season. As exposed larvae were used in connection with these experiments, the results can not be compared with those of the subsequent two winters.

On August 27 there was but a low degree of larval resistance. This was as expected, as insect activity was at its height and an occurrence of temperatures comparable to those employed in the laboratory would be rare indeed for that season of the year. As decreasing fall temperatures occurred, it will be seen that larval resistance increased until midwinter, when the maximum cold-hardiness prevailed.

On October 10 the data are sufficiently out of line to warrant some explanation, as larvae used in this test and previously on September 24 were from the same source. As moderate, rainy weather prevailed on October 10, it is possible that although the larvae were sluggishly active when collected, their resistance was still further reduced during transportation from the field to the laboratory. This occurrence illustrates the degree to which the cold-hardiness of mountain pine beetle larvae will fluctuate under changed climatic conditions.

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\* Relation of Unseasonal Temperatures to Bark Beetle Mortality.  
James C. Evenden, March 26, 1940. Unpublished report.



Throughout these experiments an outstanding difference in the cold-hardiness of individual larvae is portrayed. This variation is not entirely one of larval development, for although prepupal larvae show the greatest resistance, there is a distinct variation in larvae of the same instar. A number of factors may play an important part in determining individual larval resistance. Under no known conditions do all larvae, even in the same tree, reach the same degree of cold-hardiness, although their resistance would seem to change proportionately. This condition is definitely shown in all tests by the occurrence of some mortality at temperatures considerably higher than that required to produce 100 percent mortality.

Extreme low temperatures were employed during the January 12 experiment. The larvae which temporarily recovered from these exposures were placed in artificially constructed cells in the phloem of fresh white pine wood. On January 22, a number of larvae from the  $-61^{\circ}$  F. exposure had done some feeding; however, they soon became inactive and died. Larvae from the  $-78^{\circ}$  F. exposure lived until January 22, but did not feed. Although not shown on the chart, larvae were exposed to  $-116^{\circ}$  F., and although 68 percent of them recovered and became active, they all died prior to January 22. Although these larvae shown an apparent recovery, their ultimate death when exposed to such extreme temperatures would seem to be assured.



### WINTER TEMPERATURES OF THE PAST THREE SEASONS

During the winters of 1938-39 and 1940-41 overwintering mountain pine beetle larvae in white pine developed a high degree of cold-hardiness. Unfortunately the difference in techniques employed does not permit a comparison of results, but it is believed that a higher degree of larval resistance was attained in 1938-39 than during the past season. On the other hand, during the winter of 1939-40 the degree of cold-hardiness developed by overwintering mountain pine beetle larvae was much lower than during the subsequent or previous season. As during the last two seasons the same technique of operation was employed, a comparison of the measured larval resistance is possible:

<u>Winter</u>	<u>Degree of cold-hardiness</u>	<u>Month</u>
1938-39	-20° F.	January
1940-41	-40° F.	January

Obviously there is a reason for this seasonal difference in cold-hardiness. A comparison of charts II, III and IV offers a plausible explanation which supports the contention that the severity or tempering influences exerted by fall temperatures are responsible. Charts II and IV show that with the exception of a few days of extreme cold in February 1939 the two seasons were somewhat equal in severity. Although fall temperatures of 1940 were a trifle more severe than those of 1938, they are sufficiently close for one to assume that fairly comparable degrees of cold-hardiness would prevail, which is an assumption supported by the data obtained from the experiments. In further support of this position, chart III shows higher fall temperatures than for the other two seasons, which explains the low degree of larval resistance developed during that season.

Regardless of this variation, it was apparent that the lower resistance was still sufficient to withstand the near zero temperatures which occurred during January 1940.

### CONCLUSIONS

The data obtained in connection with low-temperature experiments conducted at this laboratory show that under normal weather conditions sufficient cold-hardiness is developed by overwintering larvae of the mountain pine beetle to withstand the minimum seasonal temperatures. These data also show the potential importance of unseasonal low temperatures as a direct cause of abnormal brood mortality. Such unseasonal low temperatures could occur in the fall prior to development of an adequate cold-hardiness, or in the spring after overwintering resistance had been broken by activity temperatures. Periods of excessive warm weather during the winter which are followed by normal temperatures could break larval resistance to such an extent that brood mortality would follow. Excessive brood mortality often found under the thin bark of the upper bole of infested trees can perhaps be explained by the occurrence of such a condition. As a result of these tests the potential value of low temperatures as a means of reducing bark beetle populations is recognized, as well as an evaluation of climatic conditions responsible.

CHAPTER I  
SEASONAL RESISTANCE OF MOUNTAIN PINE BEETLE LARVAE TO LOW TEMPERATURES  
WHITE PINE  
Larval Mortality Shown in percent

Date	+20	+10	0	-10	-20	-30	-40	-50	-60	-70
1938										
Aug. 27	0	20	100							
Sept. 9	0	4	8	88	96	100				
Sept. 24	0	2	24	92	90	100				
Oct. 10	0	60	88	86	96	100				
Oct. 24	0	2	10	34	32	56	76	100		
Nov. 7			0	2	16	52	84	88	92	96
Nov. 21			0	6	8	10	10	14	18	22 44
Nov. 28				0	2	4	4	10	8	14 10
Dec. 12						14	8	12	8	10 22 24 30
1939										
Jan. 1					0	2	4	14	14	18 26 26
Jan. 12									8	14
March 6						10	10	34	28	
March 20			0	6	8	32	58	60	70	
April 6		8	36	74	84	98	96	98	98	100
April 17	0	34	68	76	76	92	100			
May 1	6	22	82	100						
May 15	0	8	26	62	90	96				



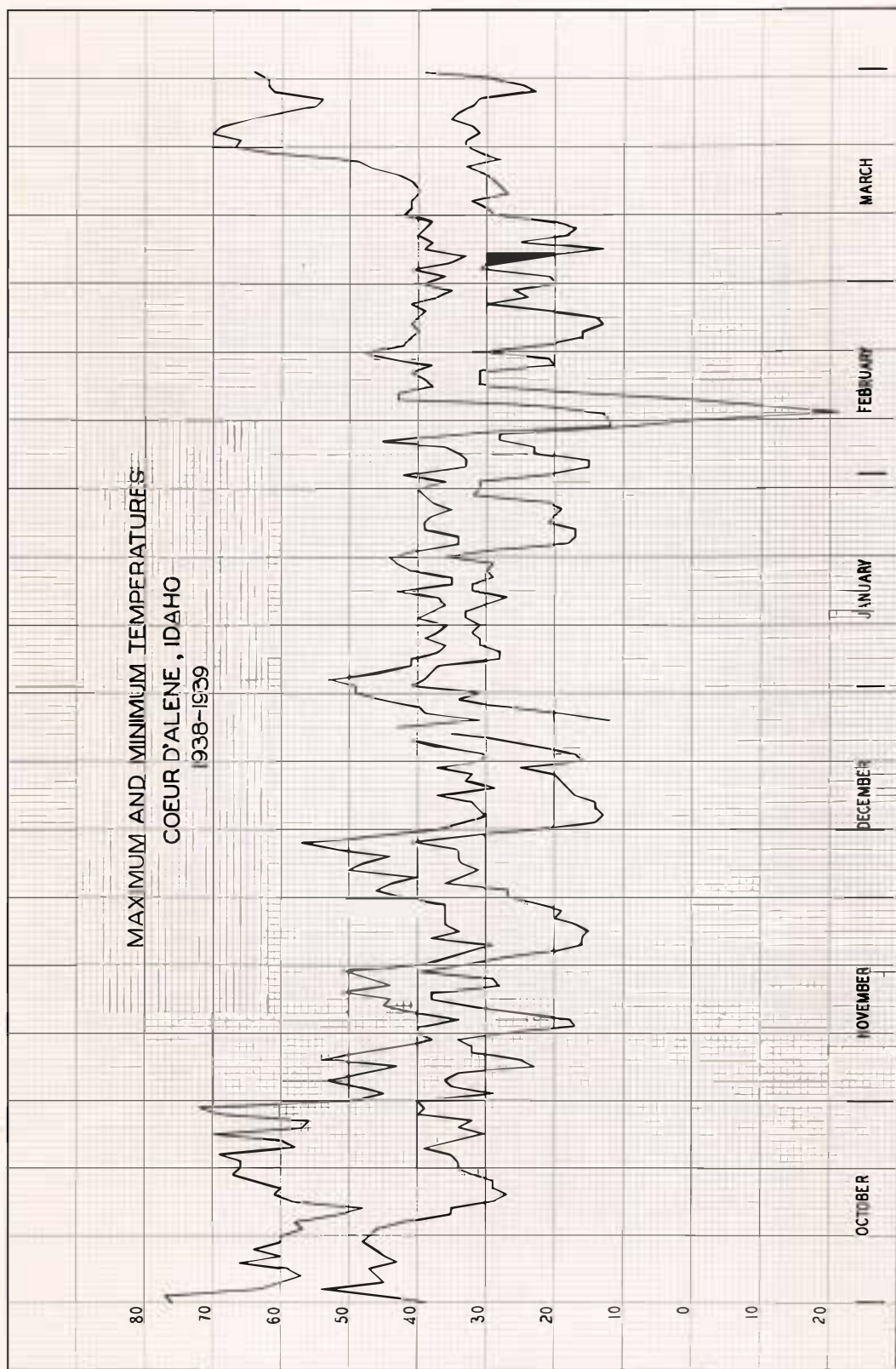


CHART II



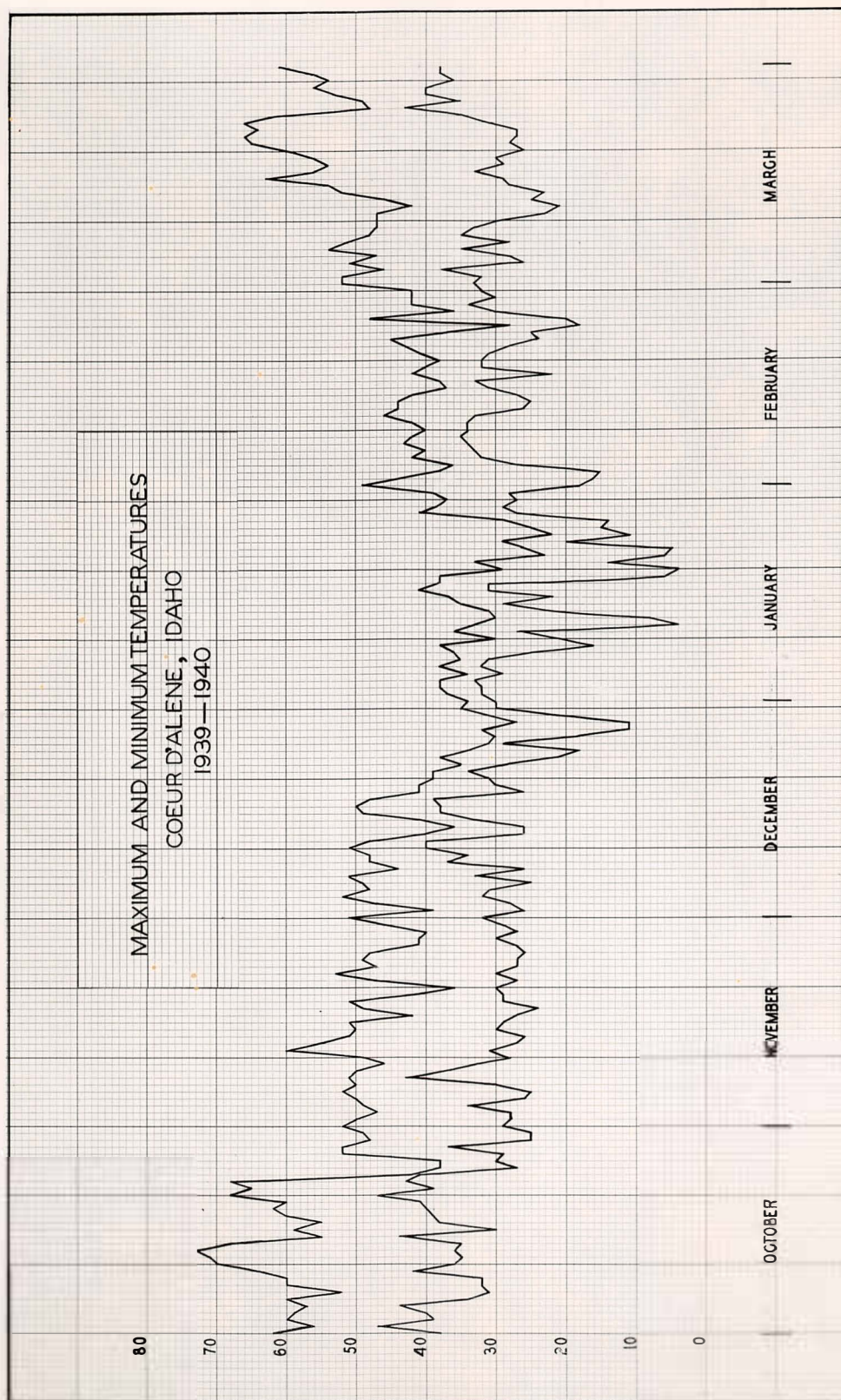


CHART III

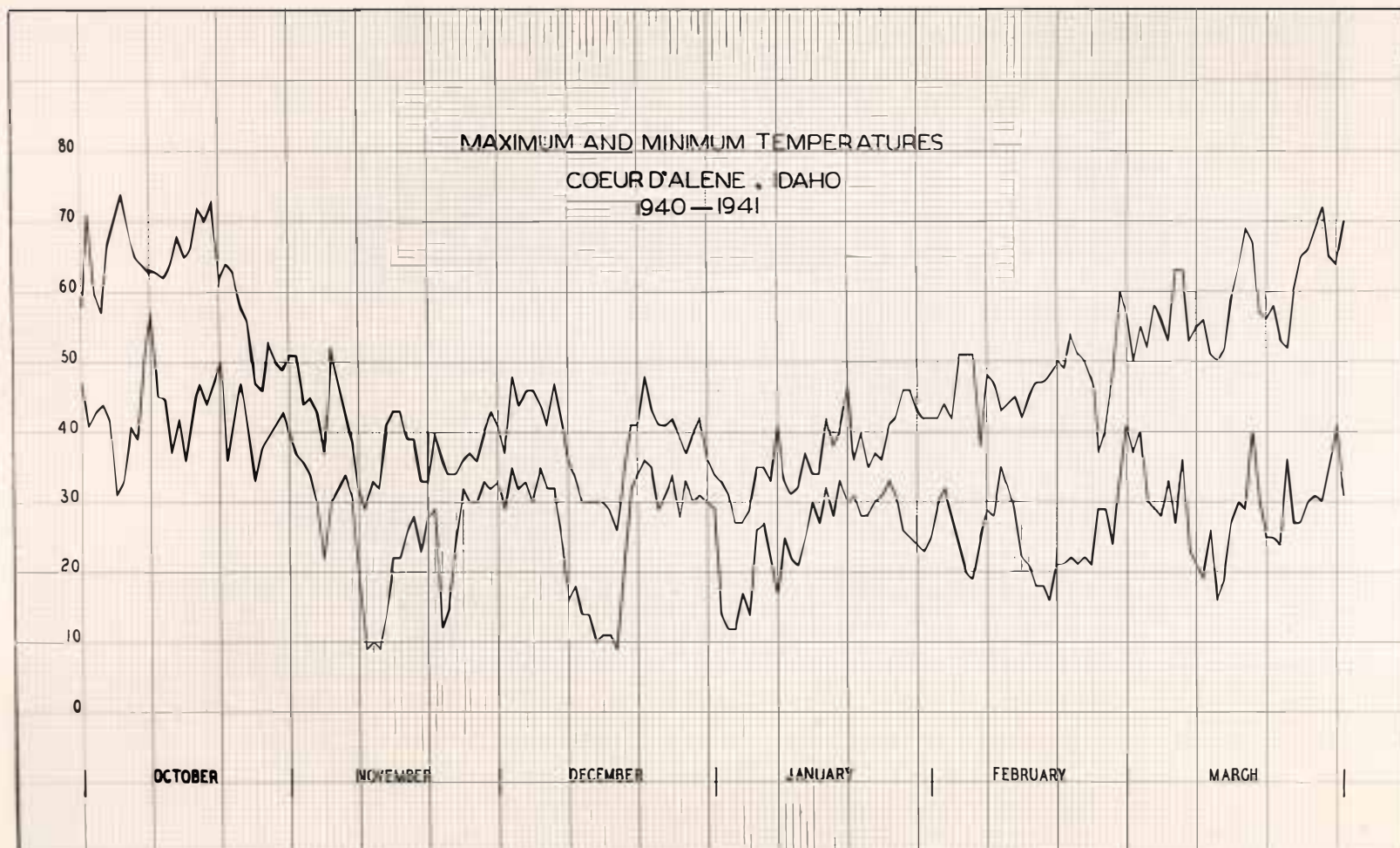


CHART IV